WEST Search History

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DB=US	SPT; PLUR=YES; OP=OR		
L9	hydrophilic and 11	0	L9
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L7	oil and 11	0	L7
L6	phaseolin and 11	0	L6
L5	seed adj specific adj promoter and 11	0	L5
L4	seed-specific adj promoter and 11	0	L4
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1. <u>6288303</u> . 25 Jun 98; 11 Sep 01. Rice .betaglucanase enzymes and genes. Rodriguez; Raymond L 800/287; 435/320.1 435/468 435/69.8 536/23.6 536/24.1 800/298 800/320 800/320.2. C12N015/29 C12N015/56 C12N015/82 A01H005/00 A01H005/10.
2. <u>6183739</u> . 26 Mar 99; 06 Feb 01. Phospholipases in animal feed. Beudeker; Robert Franciscus, et al. 424/94.6; 424/442 426/635 435/197 800/298. A01H005/00 A23K001/00 A23K001/14 A61K038/46 C12N009/18.
3. 6017530. 15 May 96; 25 Jan 00. Phospholipases in animal feed. Beudeker; Robert Franciscus, et al. 424/94.6; 424/442 435/197. A61K038/46 A23K001/165 C12N009/18.
4. <u>5994628</u> . 02 Jun 95; 30 Nov 99. Process for protein production in plants. Rodriguez; Raymond L. 800/298; 435/320.1 435/468 435/69.1 435/69.8 536/23.5 536/23.6 536/24.1. C12N015/29 C12N015/82 C12N015/12 A01H005/00.
5. <u>5948682</u> . 25 Apr 97; 07 Sep 99. Preparation of heterologous proteins on oil bodies. Moloney; Maurice M. 435/483; 435/183 435/214 435/219 435/254.2 435/254.21 435/320.1 435/477 435/69.1 435/69.2 435/69.4 435/69.52 435/69.6 435/69.7 435/69.8 435/70.1 435/71.1 536/23.2 536/23.4 536/23.52 536/23.6 536/24.1. C12N015/81 C12N015/29 C12N015/62 C12N015/12.
6. <u>5889189</u> . 02 Jun 95; 30 Mar 99. Process for protein production in plants. Rodriguez; Raymond L. 800/320; 435/320.1 435/69.1 435/69.8 536/23.5 536/23.6 536/24.1 800/288 800/320.1 800/320.2 800/320.3. C12N015/29 C12N015/82 C12N015/12 A01H005/00.
7. <u>5888789</u> . 02 Jun 95; 30 Mar 99. Process for protein production in plants. Rodriguez; Raymond L. 435/69.1; 435/320.1 435/419 435/420 435/431 435/468 435/69.8 435/70.1 530/412 536/23.6 536/24.1 800/278 800/288. C12N015/29 C12N015/82 C12N015/09 A01H004/00.
8. <u>5714474</u> . 02 Apr 96; 03 Feb 98. Production of enzymes in seeds and their use. Van Ooijen; Albert J. J., et al. 514/44; 119/174 426/20 426/21 426/629 426/630 426/635 435/183 435/196 435/202 435/69.1 514/2. A61K048/00 A61K038/43 A01H005/10 C12N015/82.
9. <u>5693506</u> . 16 Nov 93; 02 Dec 97. Process for protein production in plants. Rodriguez; Raymond L 536/23.2; 435/204 435/69.8 536/23.4 536/23.51 536/23.53 536/23.6 536/23.72 536/24.1 536/25.3. C12N015/09 C12N015/29 C07H021/04 A01H001/00 A01H001/04.
10. <u>5650554</u> . 30 Dec 94; 22 Jul 97. Oil-body proteins as carriers of high-value peptides in plants. Moloney; Maurice. 800/288; 435/183 435/320.1 435/418 435/419 435/69.1 435/69.2 435/69.52 435/69.6 435/69.7 435/69.8 435/70.1 435/71.1 536/23.2 536/23.4 536/23.52 536/23.6 536/24.1 800/298 800/301 800/302. A01H005/00 C12N015/29 C12N015/82 C12N005/04.
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L8: Entry 1 of 1

File: USPT

Dec 2, 1997

DOCUMENT-IDENTIFIER: US 5693506 A

TITLE: Process for protein production in plants

<u>US PATENT NO.</u> (1): 5693506

Brief Summary Text (16):

A two-step process for producing recombinant proteins in cereal seeds and cell cultures derived from cereal plants, is provided. Genes encoding recombinant proteins are inserted into two separate expression constructs. One construct uses metabolically regulated promoters to achieve expression of the recombinant protein in plant cell culture or, during germination (i.e., malting) of transgenic seed. The other expression construct uses a hormonally regulated promoter to achieve expression of the recombinant protein in the malted seed. Both constructs utilize additional regulatory DNA sequences that permit the target protein to be secreted extracellularly. Cells or tissues derived from cereal plants can be transformed singly or together (i.e., co-transformation) with the expression constructs. Transgenic callus tissue derived from these transformation events are used to express the recombinant protein in cell culture. The resulting transgenic calli can also be used to regenerate whole transgenic plants that produce viable seeds. Using this two-step process, the recombinant protein can be recovered and purified first, from the cell culture medium and second, from the mash (seed protein extract) from malted transgenic seeds. Expression of recombinant proteins in the malting system can be further maximized by modifying the malting process to accommodate dehulled, de-embryonated seeds and isolated embryos.

Detailed Description Text (13):

Cells or tissues or derived from cereal plants can be transformed singly or together (i.e., co-transformation) with the expression constructs. Once integrated into the plant genome, the recombinant protein can be recovered and purified first, from the cell culture medium and second, from the mash (seed protein extract) from malted transgenic seeds. The principle of using different cereal a-amylase promoters to express a recombinant protein in plant cell culture and in transgenic seeds is illustrated in FIGS. 1A and 1B. In this figure, the sugar-repressible promoter for the rice .alpha.-amylase gene, RAmy3D, and the gibberellic acid-induced promoter for the RAmylA gene, were used to express the bacterial reporter gene, qusA, in rice. The gusA gene encodes the enzyme, beta-glucuronidase (GUS), that produces a blue chromophore in tissues expressing the gene. This chromophore can be easily detected using a histochemical staining method. As can been seen in this figure, the product of gusA is repressed in rice cells when the culture medium contains 3% sugar. In transgenic rice seeds containing the RAmylA promoter/GUS fusion, the blue chromophore increases up to six days of germination. Using this two-step expression system, cereal species such as rice, corn, wheat, oats, rye, barley and various grasses can be genetically engineered to express a wide range of recombinant proteins in either or both stages. By combining this unique technology with well-established production methods (e.g., plant cell fermentation, crop cultivation, malting, and product recovery), recombinant protein can be efficiently and economically produced for the biopharmaceutical, industrial processing, animal health and bioremediation industries. The fact that this expression system does not require the use of genetic elements derived from animal or plant pathogens should facilitate regulatory acceptance.

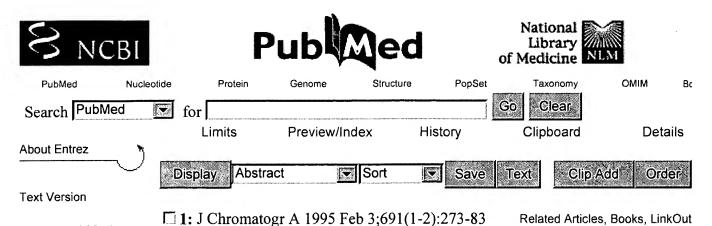
Detailed Description Text (17):

By using a number of standard procedures, one of skill can identify suitable promoters and signal sequences for use in this invention in other species of plants. While the gene can be amplified directly from a mRNA extract using PCR, the first step is

- 47. The process of claim 46 wherein said filamentous fungus is Mucor or Humicola.
- 48. The process of claim 47 wherein said gene from said Humicola comprises a glucoamylase gene.
- 49. The process of claim 47 wherein said gene from said 5 Mucor comprises a carboxyl protease gene.
- 50. The process of claims 45 or 46 wherein said culturing is carried out in a culture medium comprising utilizable carbon, nitrogen and phosphate sources, surfactant and trace elements.
- 51. The process of claims 45 or 46 further comprising the step of isolating said secreted heterologous polypeptide.
- 52. The process of claims 45 or 46 wherein said heterologous polypeptide is biochemically active.
- 53. The process of claims 45 or 46 wherein said heter- 15 ologous polypeptide comprises a mammalian polypeptide.
- 54. The process of claim 53 wherein said mammalian polypeptide comprises chymosin or prochymosin.
- 55. The process of claims 45 or 46 wherein said heterologous polypeptide comprises a polypetide from a filamen- 20 tous fungus other than said Aspergillus host.
- 56. The process of claims or 46 wherein said heterologous polypeptide comprises an enzyme.
- 57. The process of claims 45 or 46 wherein said enzyme is selected from the group consisting of chymosin, prochy-25 mosin, Aspergillus niger glucoamylase, Humicola grisea glucoamylase and Mucor miehei carboxyl protease.
- 58. The process of claims 45 or 46 wherein said signal peptide is from a polypeptide secreted from a filamentous fungus.
- 59. The process of claims 45 or 46 wherein said signal peptide is from a secreted polypeptide selected from the group consisting of Aspergillus glucoamylase, Humicola glucoamylase, and Mucor carboxyi protease.
- 60. The process of claims 45 or 46 wherein said signal 35 peptide is from a source other than a filamentous fungus.
- 61. The process of claims 45 or 46 wherein said signal peptide comprises the signal peptide from a secreted mammalian polypeptide.
- 62. The process of claims 45 or 46 where said mammalian 40 polypeptide comprises preprochymosin.
- 63. The process of claims 45 or 46 wherein said vector further comprises DNA encoding a selection characteristic expressible in said Aspergillus host.
- 64. The process of claims 45 or 46 wherein said election 45 characteristic is selected from the group consisting of the acetamidase gene, pgr4, argB and trpC.
- 65. The process of claims 45 or 46 wherein said Aspergillus host is selected from the group consisting of Aspergillus

- niger, Aspergillus oryzae, Aspergillus awamori and Aspergillus nidulans.
- 66. The process of claims 38 or 41 wherein said culturing is carried out in a culture medium comprising utilizable carbon, nitrogen and phosphate sources, surfactant and trace elements.
- 67. The process of claims 38 or 41 further comprising the step of isolating said secreted heterologous polypeptide.
- 68. The process of claims 38 or 41 wherein said heter-10 ologous polypeptide is biochemically active.
 - 69. The process of claims 38 or 41 wherein said heterologous polypeptide comprises a mammalian polypeptide.
 - 70. The process of claim 69 wherein said mammalian polypeptide comprises chymosin or prochymosin.
 - 71. The process of claims 38 or 41 wherein said heterologous polypeptide comprises a polypeptide from a filamentous fungus other than said Aspergillus host.
 - 72. The process of claims 38 or 41 wherein said heterologous polypeptide comprises an enzyme.
 - 73. The process of claims 38 or 41 wherein said enzyme is selected from the group consisting of chymosin, prochymosin, Aspergillus niger glucoamylase, Humicola grisea glucoamylase and Mucor miehei carboxyl protease.
 - 74. The process of claims 38 or 41 wherein said signal peptide is from a polypeptide secreted from a filamentous fungus.
 - 75. The process of claims 38 or 41 wherein said signal peptide is from a secreted polypeptide selected from the group consisting of Aspergillus glucoamylase, Humicola glucoamylast, and Mucor carboxyl protease.
 - 76. The process of claims 38 or 41 wherein said signal peptide is from a source other than a filamentous fungus.
 - 77. The process of claims 38 or 41 wherein said signal peptide comprises the signal peptide from secreted mammalian polypeptide.
 - 78. The process of claims 38 or 41 where said mammalian polypeptide comprises preprochymosin.
 - 79. The process of claims 38 or 41 wherein said vector further comprises DNA encoding a selection characteristic expressible in said Aspergillus host.
 - 80. The process of claims 38 or 41 wherein said election characteristic is selected from the group consisting of the acetamidase gene, pyr4 argB and trpC.
 - 81. The process of claims 38 or 41 wherein said Aspergillus host is selected from the group consisting of Aspergillus niger, Aspergillus oryzae, Aspergillus awamori and Aspergillus nidulans.

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High-performance liquid chromatography of amino acids, peptides and proteins. CXXXIX. Impact of operating parameters in large-scale chromatography of proteins.

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Large-scale chromatography has been playing an important role in downstream treatment processing in biotechnology. In order to improve the productivity, the throughput of the chromatographic equipment was often increased by increasing the flow-rate and/or by increasing the column sample loading. This paper reports the results of a study on the impact of these and other operating parameters in affinity and ion-exchange chromatographic columns when used for protein purification. A sectional model was developed to predict protein adsorption processes in a packed column. The formulations of this mathematical model are presented in the Appendix. The present study was carried out with computer simulation based on this model and using data obtained from laboratory-scale columns. This model can simulate both the adsorption and washing stages of the protein purification process for both porous and non-porous particles. The effects of changing operating parameters were simulated and contour plots were generated for the easy identification of these effects. It was shown that both flow-rate and column loading can have a considerable impact on the processing rate and the yield of the column. As for the column capacity utilization, the impact of changing flow-rate is not significant at column loading of less than 80% in the test case. It was suggest that the present investigation provides a systematic predictive strategy which will greatly reduce the need for expensive, labour-intensive and time-consuming experimental work during process scale-up.

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